CLAIMS

WHAT IS CLAIMED IS:

1. A composition comprising an orthogonal aminoacyl-tRNA synthetase (O-RS), wherein the O-RS preferentially aminoacylates an O-tRNA with an efficiency of at least 50% of the efficiency of a polypeptide comprising an amino acid sequence of SEQ ID NO.: 18 with a keto amino acid.

2. The composition of claim 1, wherein the O-RS comprises an amino acid sequence comprising any one of SEQ ID NO.: 18-20, or a conservative variation thereof.

3. The composition of claim 1, wherein the O-RS is derived from a *Methonococcus jannaschii.*

4. The composition of claim 1, comprising a cell.

5. The composition of claim 4, wherein the cell is an *E. coli* cell.

6. The composition of claim 1, comprising a translation system.

7. The composition of claim 1, further comprising an O-tRNA.

8. The composition of claim 7, wherein the O-tRNA comprises or is encoded by a polynucleotide sequence of SEQ ID NO.:21.

9. A cell comprising a translation system, wherein the translation system comprises:
an orthogonal -tRNA (O-tRNA);
an orthogonal aminoacyl-tRNA synthetase (O-RS); and,
a keto amino acid;
wherein the O-tRNA recognizes a first selector codon, and the O-RS preferentially aminoacylates the O-tRNA with an efficiency of at least 50% of the efficiency of a polypeptide comprising an amino acid sequence of SEQ ID NO.: 18 with the first keto amino acid.

10. The cell of claim 9, wherein the O-tRNA comprises or is encoded by a polynucleotide sequence as set forth in SEQ ID NO.: 21, or a complementary polynucleotide sequence thereof, and wherein the O-RS comprises an amino acid sequence comprising any one of SEQ ID NO.: 18-20, or a conservative variation thereof.

11. The cell of claim 9, wherein the keto amino acid comprises a *p*-acetyl-L-phenylalanine.

12. The cell of claim 9, wherein the cell is a non-eukaryotic cell.

13. The cell of claim 12, wherein the non-eukaryotic cell is an *E. coli* cell.

14. The cell of claim 9, further comprising a nucleic acid that comprises a polynucleotide that encodes a polypeptide of interest, wherein the polynucleotide comprises a selector codon that is recognized by the O-tRNA.

15. An *E. coli* cell, comprising:

an orthogonal tRNA (O-tRNA);

an orthogonal aminoacyl- tRNA synthetase (O-RS), wherein the O-RS preferentially aminoacylates the O-tRNA with an efficiency of at least 50% of the efficiency of a polypeptide comprising an amino acid sequence of SEQ ID NO.: 18 with a keto amino acid;

the keto amino acid; and,

a nucleic acid that comprises a polynucleotide that encodes a polypeptide of interest, wherein the polynucleotide comprises the selector codon that is recognized by the O-tRNA.

16. The *E. coli* cell of claim 15, wherein the O-tRNA comprises or is encoded by a polynucleotide sequence as set forth in SEQ ID NO.: 21, or a complementary polynucleotide sequence thereof, and wherein the O-RS comprises an amino acid sequence comprising any one of SEQ ID NO.: 18-20, or a conservative variation thereof.

17. An artificial polypeptide comprising any one of SEQ ID NO. 18-20.

18. An artificial polynucleotide that encodes a polypeptide of claim 17.

19. A vector comprising or encoding a polynucleotide of claim 18.

20. The vector of claim 19, wherein the vector comprises a plasmid, a cosmid, a phage, or a virus.

21. The vector of claim 19, wherein the vector is an expression vector.

22. A cell comprising the vector of claim 19.

23. A method of producing a protein in a cell with a keto amino acid at a specified position, the method comprising:

growing, in an appropriate medium, the cell, where the cell comprises a nucleic acid that comprises at least one selector codon and encodes a protein; and,

providing the keto amino acid;

wherein the cell further comprises:

an orthogonal tRNA (O-tRNA) that functions in the cell and recognizes the selector codon; and,

an orthogonal aminoacyl-tRNA synthetase (O-RS) that preferentially aminoacylates the O-tRNA with an efficiency of at least 50% of the efficiency of a polypeptide comprising an amino acid sequence of SEQ ID NO.: 18 with the keto amino acid; and

incorporating the keto amino acid into the specified position in the protein during translation of the nucleic acid with the at least one selector codon, thereby producing the protein.

24. The method of claim 23, wherein the O-RS comprises an amino acid sequence which comprises any one of SEQ ID NO.: 18-20.

25. The method of claim 23, wherein the O-tRNA comprises or is encoded by a polynucleotide sequence as set forth in SEQ ID NO.: 21, or a complementary polynucleotide sequence thereof

26. The method of claim 23, wherein the cell is a non-eukaryotic cell.

27. The method of claim 26, wherein the non-eukaryotic cell is an *E. coli* cell.

28. The method of claim 23, wherein the keto amino acid is *p*-acetyl-L-phenylalanine.